STATISTICAL ANALYSIS PLAN
Protocol: 04-02

Safety and Effectiveness of the Alair® System for the Treatment of Asthma:

A Multi-center Randomized Clinical Trial (Asthma Intervention Research (AIR2) Trial)

Prepared for:

Asthmatx, Inc.

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August 21, 2008 / Version 1

Approved by:	**Signature**	**Date**
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1. INTRODUCTION

Asthma is a serious health problem throughout the world. It is one of the most common and costly diseases in the U.S. (CDC 1999) with an estimate of more than 20 million sufferers (National Center for Health Statistics 2000-2001). In 1998, nearly half of the American public said they had asthma themselves, in their household, or in their immediate family (Schulman 1998). In 2005, asthma resulted in 13.6 million unscheduled physician office visits, 1.8 million emergency department visits, 465,000 hospital admissions (average length of stay of 3 days) and more than 4,000 deaths (National Center for Health Statistics 2005). Asthma-related healthcare costs are estimated at $19.7 billion per year (National Center for Health Statistics 2005). The prevalence and mortality of the disease are on the rise, current treatments are limited, and there is no known cure. According to the Department of Health and Human Service's Strategic Plan, Action Against Asthma, which describes how HHS will tackle asthma over the next five years, HHS established as one of its four priorities to "reduce the burden of asthma for people with the disease" (DHHS Federal Asthma Research Agenda, 1999). Asthma is an often debilitating disease characterized by dyspnea, wheezing, coughing, and respiratory distress. Patients with asthma typically have hyperresponsive and often chronically inflamed airways (Cox 2004). Chronic asthma is characterized by extensive airway remodeling, with thickening of airway walls, increased mucous gland and goblet cells, increased vascularization, and hypertrophy of airway smooth muscle (ASM). Although there are many possible triggers, asthma is invariably associated with airways that narrow too easily and/or too much in response to provocative stimuli (National Institutes of Health Global Initiative for Asthma, GINA 2002). Thus, regardless of the initial trigger (e.g., allergen, irritant, infection), the cascade ends with ASM contraction with subsequent airway narrowing and airflow obstruction.

It is therefore believed that reduction in the amount of functioning airway smooth muscle will decrease bronchoconstriction. Thus, a therapy that reduces ASM mass or reduces the ability of ASM to contract has the potential to reduce bronchoconstriction and the symptoms of asthma.

2. STUDY OBJECTIVES

The objective of this randomized, double blind, sham-controlled study is to demonstrate the safety and effectiveness of the Alair® System in a population of subjects with severe asthma who are still symptomatic despite being managed on conventional therapy of high doses of inhaled corticosteroids (ICS - doses greater than 1000µg per day beclomethasone or equivalent) and long-acting β_2-agonists (LABA - doses of at least 100µg per day salmeterol or equivalent).

3. DESIGN

3.1 Overview

This is a randomized, double blind, sham-controlled study is to demonstrate the safety and effectiveness of the Alair® System in a population of subjects with severe asthma who are still symptomatic despite being managed on conventional therapy of

high doses of inhaled corticosteroids (ICS - doses greater than 1000µg per day beclomethasone or equivalent) and long-acting β_2-agonists (LABA - doses of at least 100µg per day salmeterol or equivalent).

3.2 Expected Sample Size

The goal is for a sample size of 225 after subjects who withdraw or are lost-to-follow-up. This sample size results in an expectation of 150 treatment subjects and 75 control subjects. The study will accrue 250 subjects to make sure, after lost-to-follow-up, that there is a minimum of 225 evaluable subjects. During accrual, if the blinded results show an attrition rate larger than the expected 10% the study may continue enrolling up to a maximum of 300 subjects.

3.3 Inclusion/Exclusion Criteria

3.3.1 Inclusion Criteria

1. Subject is an adult between the ages of 18 to 65 years.
2. Subject is able to read, understand, and sign a written Informed Consent to participate in the Study.
3. Subject has asthma and is taking regular maintenance medication that includes:
    a. Inhaled corticosteroid (ICS) at a dosage greater than 1000µg beclomethasone per day or equivalent, AND long acting ß2-agonist (LABA) at a dosage of ≥100µg per day Salmeterol or equivalent.
    b. Other asthma medications such as leukotriene modifiers, or anti-IgE, are acceptable (subjects on Xolair® must have been on Xolair for greater than 1 year).
    c. Oral corticosteroids (OCS) at a dosage of up to, but not greater than 10mg per day are acceptable.*
4. Subject has an AQLQ score during the Baseline Period of 6.25 or less.
5. Subject has a Pre-bronchodilator FEV_1 of greater than or equal to 60% of predicted after medication stabilization during the Baseline Period.
6. Subject has a $PC_{20} < 8$ mg/ml per methacholine inhalation test using standardized methods.**
7. Subject has at least two days of asthma symptoms during the 4-weeks of the Baseline Diary Period.
8. Subject is a non-smoker for 1 year or greater (if former smoker, less than 10 pack years total smoking history).
9. Subject is able to undergo bronchoscopy in the opinion of the investigator or per hospital guidelines.
10. Subject is willing and able to comply with the Study protocol, including requirements for taking and abstaining from medications.

** NOTE: Subjects on a dosage regimen of 20mg OCS every other day may be included as this averages out to a daily dosage of 10mg.*

*** NOTE: If a Subject cannot tolerate a 48-hour LABA withdrawal for the methacholine challenge test, the test should be performed after a 24-hour withdrawal. Allow the Subject time to recover from the failed 48-hour LABA withdrawal (re-stabilization on their LABA regimen) before proceeding to a 24-hour LABA withdrawal attempt. In any case, all subsequent methacholine tests for each Subject must be conducted using the same LABA withdrawal time regimen as at Baseline for that Subject.*

3.3.2 Exclusion Criteria

1. Subject is participating in another clinical trial within 6 weeks of the Baseline Period involving respiratory intervention that could affect the outcome measures of this Study.
2. Subject requirement during the Baseline Diary period for rescue medication use other than for prophylactic use for exercise exceeds an average of:
    a. 8 puffs per day of short-acting bronchodilator, or
    b. 4 puffs per day of long-acting rescue bronchodilator, or
    c. 2 nebulizer treatments per day.
3. Subject has a Post-bronchodilator FEV_1 of less than 65%.
4. Subject has 3 or more hospitalizations for exacerbations of asthma in the previous year; OR a history of life-threatening asthma, defined by past intubations for asthma, or ICU admission for asthma within the prior 24 months.
5. Subject has a history of recurrent lower respiratory tract infections requiring antibiotics (more than 3 in the past 12 months).
6. Subject has a history of recurrent oral steroid use for asthma (4 or more pulses of oral steroids in the past 12 months).
7. Subject has a known sensitivity to medications required to perform bronchoscopy (such as lidocaine, atropine and benzodiazepines).
8. Subject has known systemic hypersensitivity or contraindication to Methacholine chloride or other parasympathomimetic agents.
9. Subject is undergoing immunosuppressant therapy (e.g., methotrexate).
10. Subject uses systemic β-adrenergic blocking agents.
11. Subject is on anticoagulant medication.
12. Subject is an insulin-dependent diabetic.
13. Subject is pregnant or a nursing mother, or has plans to become pregnant within the next year.
14. Subject has other respiratory diseases including emphysema, cystic fibrosis, vocal cord dysfunction, mechanical upper airway obstruction, obstructive sleep apnea, Churg-Strauss syndrome, cardiac dysfunction, and allergic bronchopulmonary aspergillosis (total IgE of >1000 Units/mL with positive specific IgE to aspergillus and evidence of central bronchiectasis).
15. Subject has segmental atelectasis, lobar consolidation, significant or unstable pulmonary infiltrate, or pneumothorax, confirmed on x-ray.
16. Subject has interstitial lung disease.
17. Subject has chronic sinus disease as defined by 5 or more episodes of sinusitis in past 12 months or continuous symptoms of sinus infection

(purulent discharge) and significant change in nasal steroid dosage in last 6 weeks.

18. Subject has uncontrolled gastro-esophageal reflux disease as defined by a significant increase in therapy in last 6 weeks.
19. Subject has significant co-morbid illness such as cancer, renal failure, liver disease, or cerebral vascular disease.
20. Subject has a history of epilepsy.
21. Subject currently has clinically significant cardiovascular disease, including myocardial infarction, angina, cardiac dysrhythmia, conduction defect, cardiomyopathy, or stroke.
22. Subject has bleeding diathesis, platelet dysfunction, and thrombocytopenia with platelet count less than 125,000/mm^2 or known coagulopathy (INR > 1.5).
23. Subject has uncontrolled hypertension (>200mm Hg systolic or >100mm Hg diastolic pressure).
24. Subject has a known aortic aneurysm.
25. Subject has an implanted electrical stimulation device (e.g., a pacemaker, cardiac defibrillator, or deep nerve or deep brain stimulator).
26. Subject has a psychiatric disorder that in the judgment of the investigator could interfere with provision of informed consent, completion of tests, therapy, or follow-up.
27. Subject has any other medical condition that would make them inappropriate for Study participation, in the Investigator's opinion.

3.5 Test Device

The device being evaluated in this clinical study is the Alair System (Asthmatx, Inc., Sunnyvale, CA).

4 STUDY SCHEMA

Procedure / Assessment	Baseline Period	Random-ization & 1st Bronch.	F/U Visit 1	2nd Bronch.	F/U Visit 2	3rd Bronch.	F/U Visit 3	6-wk F/U Visit[2]	12-wk F/U Visit	6-mo F/U Visit	9-mo F/U Visit	12-mo F/U Visit part 1 ON LABA[2]	12-mo F/U Visit part 2 OFF LABA
AQLQ (Juniper)	✓								✓	✓	✓	✓	✓
ACQ (Juniper)	✓								✓	✓		✓	✓
EQ5D Health Form	✓												
Medical History	✓												
Spirometry	✓1	✓1	✓1	✓1	✓1	✓1	✓1	✓1	✓1	✓1		✓1	✓1
Methacholine Challenge	✓1								✓1	✓1		✓1	
Examination of Daily Diary	✓		✓		✓		✓	✓	✓	✓		✓	✓
Physical Examination – to include SpO2	✓		✓		✓		✓	✓	✓	✓		✓	✓
Review of Asthma Symptoms, Exacerbations, Medications	✓		✓		✓		✓	✓	✓	✓	✓	✓	✓
Pregnancy Testing	✓	✓		✓		✓			✓	✓		✓	
Resting EKG	✓												
Chest X-Ray – Lateral & PA	✓												
High Resolution CT Scan	✓3											✓4	
Hematology, Coagulation, Blood Chemistry	✓												
Lung Volumes	✓											✓	
Bronchoscopy		✓		✓		✓							
Blinding Assessment Questionnaire		✓5		✓5		✓5			✓5	✓5	✓5	✓5	✓5

[1] These tests must be done following specific medication guidelines described in the protocol.
[2] Four weeks prior to these visits, schedule an "office visit" to remind Subjects to keep an accurate Daily Diary. No testing is done during this office visit.
[3] All Subjects.
[4] First 150 Subjects enrolled in the Study.
[5] Done at this time point for the Subject AND the Assessment Team members present.

	Unblinded Bronchoscopy Team

	Blinded Assessment Team

5 EFFICACY AND SAFETY ENDPOINTS

5.1 Efficacy and Safety Endpoints

5.1.1 Primary Efficacy Endpoints

The primary efficacy endpoint will be the difference between Study groups in the change in Asthma Quality of Life Questionnaire (AQLQ) score from Baseline and the average of 6-, 9-, and 12-month follow-up visits (6-, 9-, and 12-months after last bronchoscopy session).
AQLQ score is a numeric score on a 7-point scale. A higher AQLQ score represents better Quality of Life. The average of the 6-, 9-, and 12-month differences in the AQLQ score are referred to as the "Integrated AQLQ score".

5.1.2 Secondary Efficacy Endpoints

- Difference between Study groups in "Absolute Change from Baseline" in percent Symptom Free Days at 6 and 12 months (6- and 12-months after last bronchoscopy session).
- Symptom score: Difference between Study groups in Change between Baseline and 6-, and 12-month Follow-Up Visits (6- and 12-months after the last bronchoscopy session).
- Morning Peak Expiratory Flow (amPEF): Difference between Study groups in Change between Baseline and 6- and 12-Month Follow-up Visits (6- and 12-months after the last bronchoscopy session).
- Asthma Quality of Life Questionnaire (AQLQ) score: Difference between Study groups in Change between Baseline and 6- Month Follow-up Visit, Baseline and 9-Month Follow-up Visit, and Baseline and 12-Month Follow-up Visit (6-, 9-, and12-months after the last bronchoscopy session). Subset evaluation with restricted Baseline AQLQ.
- Individual Domain scores from Asthma Quality of Life Questionnaire (AQLQ): Difference between Study groups in Change between Baseline and 6- Month Follow-up Visit, Baseline and 9-Month Follow-up Visit, and Baseline and 12-Month Follow-up Visit (6-, 9-, and 12-months after the last bronchoscopy session).
- Asthma Control Questionnaire (ACQ) score: Difference between Study groups in Change between Baseline and 6- and 12-Month Follow-up Visits (6- and 12-months after the last bronchoscopy session).
- Number of puffs of rescue medication used: Difference between Study groups in Change in average number of puffs per week between Baseline and 6- and 12-Month Follow-up Visits (6- and 12-months after the last bronchoscopy session).
- Percent of Days rescue medication was used: Difference in Study groups in Change in percentage of days rescue medication was used between Baseline and 6- and 12-Month Follow-up Visits (6- and 12-months after the last bronchoscopy session).

- Forced Expiratory Volume in one second (FEV_1): Difference between Study groups in Change in FEV_1 between Baseline and 6- and 12-month Follow-Up Visits (6- and 12-months after the last bronchoscopy session).

5.1.3 Other Efficacy Endpoints

- Evening peak expiratory flow (pm PEF).
- Forced Vital Capacity (FVC).
- Methacholine PC_{20}.
- Nighttime awakenings for asthma.
- Severe asthma exacerbations
  o Incidence of severe asthma exacerbations requiring systemic steroids.
  o Number/Percentage of subjects with asthma exacerbations requiring systemic steroids.
  o Time to first exacerbation.
- Mild asthma exacerbations.
  o Incidence of mild asthma exacerbations.
  o Number/Percentage of subjects with asthma exacerbations.
  o Time to first exacerbation.
- Percent Exacerbation Qualifying Days
- Change in maintenance asthma medications.
- Percent of days that work, school, or other daily activities were affected by asthma symptoms.
- Number of subjects that withdraw from Study due to worsening of asthma.
- Change between ON-LABA at Baseline and OFF-LABA at 12-Months of key parameters.

5.2 Safety Endpoints

Safety will be evaluated with summary of adverse events.

6 STATISTICAL METHODS

6.1 Pooling of Data for Analyses

The clinical study will be conducted under a common protocol for each investigational site with the intention of pooling the data for analysis. Every effort will be made to promote consistency in study execution at each investigational site. The analysis of homogeneity will be conducted under the assumption that each investigator will have enrolled a minimum of 6 ITT subjects in the Alair treatment group and 3 subjects in the Sham treatment group. In the event that there are too few subjects in either arm for an investigator, then this investigator's data will be combined to achieve the desired sample size minimum per arm.

The process will combine the data for various geographical regions, i.e. United States sites, Australian sites, and etc. The combining of investigator's data within a region will be accomplished by taking the investigator with the smallest enrollment and combining it with the investigator with the largest. If there is a further need to combine data, then the data of the investigator with the second smallest enrollment

will be combined with the investigator's data which had the second largest enrollment, and so on. This process will continue for all investigators who did not enroll the minimum stated above. The process of combining investigator data that have insufficient subjects per arm will result in redefining the groups of investigators for the purposes of statistical analyses. These combined groups will be referred to as "analysis centers" in the statistical analyses based on an ANCOVA.

The consistency of treatment response will be investigated across the analysis centers subsequent to combining the data as described above. Statistical tests will be conducted to identify if there are extreme analysis centers that could affect the interpretation of common statistical and clinical conclusions. An analysis center by treatment interaction will be included in the primary variable analysis to test for parallel treatment effect at an alpha level of 0.05. The Integrated Change from Baseline in AQLQ will be analyzed with an ANCOVA with factors of treatment, analysis center, and treatment by analysis center interaction and the Baseline AQLQ as a covariate. Further examination will follow if the interaction effect from that ANCOVA is significant. In the event that the ANCOVA interaction p-value is less than or equal to 0.05, a sensitivity analysis that excludes analysis centers with extreme efficacy results will be performed to determine the robustness of the treatment effect. On the other hand, if the outcome of the interaction effect has a p-value greater than 0.05, then the conclusions from the pooled data will be considered to be free of the impact of extreme analysis centers.

The first step in conducting a sensitivity analysis is to identify the extreme analysis center or centers that contribute to the statistical significance of the interaction effect. The process involves submitting subsets of analysis centers to the ANCOVA and observing the interaction p-value for the subset. Subsets with p-values greater than 0.05 will be considered homogeneous.

The search for an extreme analysis center begins by analyzing all subsets that can be created by excluding one analysis center. If one or more of the subsets result in an interaction p-value greater than or equal to 0.05, then the analysis center excluded from the subset with the largest p-value will be deemed to be the extreme analysis center.

If all subset p-values are less than or equal to 0.05, then the process will analyze all subsets that can be created by excluding two analysis centers. If one or more of these subsets generates p-values larger than 0.05, then the analysis centers excluded from the subset with the largest p-value will be deemed the extreme analysis centers. Thus, the process of identifying the extreme analysis centers will continue in a stepwise manner by first excluding one, then two, then three, etc., analysis centers until the p-value exceeds 0.05.

Once the extreme analysis center or centers have been identified, then the treatment p-values of the remaining analysis centers will be computed. Inferences will be drawn from the treatment p-value, as well as any pertinent observations

regarding the extreme analysis center or centers. Additionally, it is noted that this process excludes subjects from the analysis in a non-random manner and has an unpredictable impact on the power of the treatment effect test. In the event that the treatment effect of the remaining subset is not statistically significant, due consideration of the post-hoc aspects of the process will be given when the results are interpreted. Conclusions will be presented by the sponsor as appropriate to the findings of the sensitivity analysis.

6.2 Statistical Methods

A Bayesian approach to statistical analysis will be used in this pivotal study. Bayesian statistical analysis will be used for the primary analysis as well as all secondary analyses and for the analyses of adverse events. For the primary outcome the posterior probability of superiority will be calculated. Superiority will be concluded when this probability is larger than 96.4%. See Appendix A for the Bayesian approach for the primary outcome.

Summary tables (descriptive statistics and/or frequency tables) will be provided for all baseline variables, efficacy variables, and safety variables, as appropriate. Continuous variables will be summarized with descriptive statistics (n, mean, standard deviation, range, and median). Ninety-five (95) percent confidence highest probability density (HPD) credible intervals may also be presented, as appropriate. Frequency counts and percentage of subjects within each category will be provided for categorical data.

6.3 Populations

Efficacy analyses will be performed for both the intent-to-treat (ITT) population and per-protocol (PP) population. The ITT population will consist of all randomized subjects who have been administered at least one bronchoscopy. The PP population will exclude all subjects in the ITT population who meet any of the following criteria:

- Have taken any interfering concomitant medications.
    - Addition of oral corticosteroids for non-respiratory condition, with a start date after their baseline testing and no stop date.
- Have undergone other interfering treatments.
- Did not attend one of the 6-, 9-, 12-month visits, with the exception of a discontinuation from the Study due to an adverse event related to Study treatment.
- Have missed one or more bronchoscopies.

Safety analyses will be performed on the safety population, which will be comprised of all randomized subjects who have been administered at least one bronchoscopy. The safety population is equivalent to the ITT population.

6.4 Statistical Analyses

6.4.1 Demographic and Baseline Characteristics

Demographic data including, but not limited to age, race, gender and ethnicity and baseline characteristics including, but not limited to amPEF, pmPEF, FEV_1, methacholine PC_{20}, Symptom Free Days (SFD), AQLQ score, ACQ score, and asthma medication requirements will be reported for each subject. Comparisons between the treatment groups will be conducted to assess the degree to which comparability of randomization was achieved.

6.4.2 Primary Efficacy Analyses

The primary efficacy analysis will be performed using Bayesian methods. The analysis will include baseline AQLQ as a covariate. The methods can be found in Appendix A.

6.4.3 Secondary Analyses

All secondary AQLQ analyses will include baseline AQLQ as a covariate. Secondary endpoints will be summarized with descriptive statistics. Continuous variables will be summarized with n, mean, standard deviation, and range. Frequency counts and percentage of subjects within each category will be provided for categorical data. All missing data for secondary endpoints will be imputed using Last Observation Carrier Forward (LOCF). Bayesian methods will be used to analyze secondary endpoints, as described in Appendix A. A repeated measures analysis of covariance including terms for study treatment group, time-by-treatment group interaction, and baseline AQLQ as a covariate to evaluate the AQLQ changes from baseline at 3-, 6-, 9-, and 12-months has been added as a secondary analysis.

6.4.4 Other Analyses

Other endpoints will be summarized with descriptive statistics. Continuous variables will be summarized with n, mean, standard deviation, and range. Frequency counts and percentage of subjects within each category will be provided for categorical data. Missing data will be imputed using LOCF for the following other endpoints:

- Evening peak expiratory flow (pm PEF).
- Forced Vital Capacity (FVC).
- Methacholine PC_{20}.
- Nighttime awakenings for asthma.

6.4.5 Subgroup Analyses

Subgroup analyses will be performed to identify a patient population that achieves the most benefit.

6.4.6 Safety Analyses

Safety will be evaluated by tabulations of adverse events and will be presented with descriptive statistics at Baseline and follow-up visits for each treatment group. The statistics will be organized by Treatment Phase (Bronchoscopy Periods) and Post-Treatment Phase (long-term follow-up), as appropriate.

Adverse events will be classified on the basis of MedDRA terminology and summarized for each treatment. Adverse event incidence rates will be summarized by system organ class, preferred term, and severity of the adverse event. Each subject will be counted only once within a system organ class or a preferred term by using the adverse events with the highest severity within each category. Adverse events will also be summarized by system organ class, preferred term, and relationship to procedure. Each subject will be counted only once within a system organ class or a preferred term by using the adverse events with the closest relationship to treatment within each category.

All information pertaining to adverse events noted during the Study will be listed by subject, detailing verbatim given by the investigator, preferred term, system organ class, date of onset, date of resolution, severity, and relationship to procedure. The onset of adverse events will also be shown relative (in number of days) to the day of the most recent bronchoscopy session.

A tabulation of Serious Adverse Events (SAEs) will be provided by subject within treatment groups.

The proportion of subjects in each treatment group reporting adverse events that occur in $\geq 3\%$ in either treatment group will be compared using Bayesian methods (see Appendix A). The specific system organ classes and preferred terms analyzed will be those that are reported by at least five percent of the subjects in either treatment group.

6.5 Changes to Planned Analyses

All AQLQ analyses were modified to include baseline AQLQ as covariate.

A repeated measures analysis of covariance including terms for study treatment group, time-by-treatment group interaction, and baseline AQLQ as a covariate to evaluate the AQLQ changes from baseline at 3-, 6-, 9-, and 12-months has been added as a secondary analysis.

The definition of the Intent-to-treat population was modified to include all subjects randomized and received at least one bronchoscopy.

The definition of the Safety population was modified to include all subjects randomized and received at least one bronchoscopy.

More detail was provided for the definition of per-protocol population (Section 6.3).

Bayesian imputation will be used only for the primary endpoint. Missing values for the secondary endpoints are to be imputed using LOCF. The Bayesian analyses of

secondary endpoints will be executed on LOCF data. Other endpoints described in section 6.4.4 will be imputed using LOCF.

The pooling analysis was modified from using a Bayesian Hierarchical model to the methodology in section 6.1.

The definition of a severe exacerbation is included in Section 7. The definition was modified to clarify the methods needed for programming. The baseline average morning peak flow, puffs per day, and nebulizer use are averaged from the interquartile range of the respective variable over 4 weeks.

The incidence of exacerbations is such that subjects may not have exacerbations in every time period. Given the sparseness of data, days per exacerbation and changes of this measure will not be performed.

Total days of mild and severe exacerbations was modified to present percent of exacerbation qualifying days.

The analysis in the secondary endpoint analyses of AQLQ originally proposed subset evaluation with restricted Baseline AQLQ. Since all Bayesian AQLQ analyses include baseline AQLQ as a covariate, only descriptive statistics will be provided by treatment arm for the subsets AQLQ < 2, $2 \leq$ AQLQ < 3, $3 \leq$ AQLQ < 4, $4 \leq$ AQLQ < 5, $5 \leq$ AQLQ < 6, and $6 \leq$ AQLQ.Three month summaries were added to tables for all relevant endpoints.

Summary tables of individual symptoms as collected on the diary have been added to this SAP.

FEV_1/FVC Ratio tables will be produced.

7 SCALES AND DEFINITIONS FOR CLINICAL EVALUATIONS

Protocol Definition of a Severe Exacerbation
A severe exacerbation is defined as a worsening of asthma requiring treatment with oral or intravenous corticosteroids, OR a doubling of the baseline inhaled corticosteroid dose for at least 3 days, OR any temporary increase in the dosage of oral corticosteroids for a Subject taking maintenance oral corticosteroids at Study entry.

For the purposes of severe exacerbation analysis, the following additional definition of a severe exacerbation based on Daily Diary entries will be used for the periods when subjects maintain their Daily Diary: A decrease in morning peak flow by more than 30% below the average morning peak flow during the Baseline period.

Protocol Definition of a Mild Exacerbation

A mild exacerbation is defined as 2 consecutive days when at least on of the following occurs:

- Morning Peak Expiratory Flow (PEF) falls at least 20% below the average morning PEF recorded during the Baseline period.
- Four (4) or more puffs of rescue short-acting bronchodilator are required over the average usage during the Baseline period.
- Nebulizer for short-acting bronchodilator delivery is used 3 or more times over the average usage during the Baseline period.
- Awakening at night with asthma symptoms.

Modified Definition of Severe Exacerbation for Programming Purposes
Severe Exacerbation: A severe exacerbation is defined as a worsening of asthma requiring one of the following treatments:

- Treatment with intravenous corticosteroids,
- Treatment with oral corticosteroids for any Subject not taking maintenance oral corticosteroids at Study entry OR any temporary increase in the dosage of oral corticosteroids for a Subject taking maintenance oral corticosteroids at Study entry.
- A doubling of the baseline inhaled corticosteroid dose for at least 3 days

For the purposes of severe exacerbation analysis, the following additional definition of a severe exacerbation based on Daily Diary entries will be used for the periods when subjects maintain their Daily Diary:

- A decrease in the morning peak flow by more than 30% below the average morning peak flow recorded during the Baseline period.

Additional Exacerbation Programming Decisions:

- Multiple medication records can overlap the same study day. For example, a maintenance oral corticosteroid record can span the entire study, but pulses of oral corticosteroids can occur during the study. The maintenance record may remain unchanged, and another medication record added for the additional dose. Therefore, a total daily dose of each type of medication will be determined for each day of the study to determine if there is an increase in oral corticosteroids or a doubling of baseline inhaled corticosteroid dose.
- Adverse events with increased asthma medication or steroid usage that are possibly exacerbations, but for which the adverse event name is not explicitly asthma exacerbation or asthma aggravated will be counted.
- Each day of the associated adverse event will be considered a severe exacerbation day, regardless of whether diary data is present for the days of the adverse event.
- If the adverse event for exacerbation does not have a resolution date present at the time of analysis, then the most recent visit date (scheduled or unscheduled) will be used as the resolution date.
- The length of an exacerbation will be defined as the number of days between the onset date of the exacerbation and the first two consecutive exacerbation

(mild or severe) free days. A new severe exacerbation cannot occur within 10 days of onset of a previous severe exacerbation.

- If two mild exacerbation days are followed immediately by a severe exacerbation day, this is counted as one mild and one severe exacerbation. After the start of the severe exacerbation, no events can happen for 10 days, and two consecutive exacerbation (mild or severe) free days are needed to end the severe exacerbation.

Using the definitions above, each day of the study will be classified as a severe qualifying day or a mild qualifying day. After each day is classified, the rules defined above are used to identify exacerbations. Since the end of an exacerbation requires two consecutive clear days, it is feasible for an exacerbation to last many days. Therefore, for each predetermined period for which exacerbations will be summarized, there is a chance a subject will be having an exacerbation when entering the time period. This situation will be handled differently, depending on whether the prior exacerbation is mild or severe.

- If the prior exacerbation is mild, a new mild exacerbation cannot occur until the prior exacerbation has ended. However, a mild exacerbation can turn into a severe exacerbation at any time. Therefore, for calculating mild exacerbation rates, the time period will be discounted the number of days the prior exacerbation overlaps the current time period; but for calculating severe exacerbation rates, the entire period will be used.
- If the prior exacerbation is severe, neither a new mild nor severe exacerbation can occur until the prior exacerbation has ended. Therefore, for calculating both mild and severe exacerbation rates, the time period will be discounted the number of days the prior severe exacerbation overlaps the current time period.

Time to First Severe Exacerbation

The time to first severe exacerbation will be calculated as the time from 6 weeks after the last treatment until the first severe exacerbation. Subjects that are in an exacerbation state entering this follow-up phase will be assigned a time to severe exacerbation of 0 days.

Time to First Mild or Severe Exacerbation

The time to first mild or severe exacerbation will be calculated as the time from 6 weeks after the last treatment until the first mild or severe exacerbation. Subjects that are in an exacerbation state entering this follow-up phase will be assigned a time to exacerbation of 0 days.

Definition of Symptom Free Days

Symptom Free Days will be defined as days when subject reports no cough, wheeze, breathlessness, or sputum during the daytime, and no wheeze, cough, or awakenings due to asthma symptoms during nighttime. The baseline, 3-, 6- and 12-month measurements are each recorded over a 4-week period.

Definition of AM/PM Peak Flow
AM and PM Peak Flow are defined as the average respective peak flow values recorded for each time period. The baseline, 3-, 6- and 12-month measurements are each recorded over a 4-week period.

Definition of Rescue Medication Use (Puffs)
Rescue Medication Use is defined as the average number of puffs (short acting inhaler) per 7 days. The baseline, 3-, 6- and 12-month measurements are each recorded over a 4-week period.

Definition of Percent Days Rescue Medication was Used
Percent Days Rescue Medication was Used is defined as the percent of days in each time period the subject had puffs (short acting inhaler) or nebulizer use. The baseline, 3-, 6- and 12-month measurements are each recorded over a 4-week period.

Medication Dose Conversions

Beclomethasone Equivalents	
Drug	**Conversion Factor**
Budesonide	1.25
Fluticasone	2
QVAR	2.5
Mometasone	2.5
Triamcinolone	0.5
Flunisolide	0.5
Ciclesonide	2.5

Salmeterol Equivalents	
Drug	**Conversion Factor**
Formoterol	100/24
Formoterol Fumarate	100/24

Daily Diary Symptom Scales

Wheeze During Night	
0	None
1	Slept well – slightly wheezy
2	Sleep disturbed by wheeze
3	Severe – awake most of the night

Cough During Night	
0	None
1	Slight
2	Moderate
3	Severe

Wheeze During Day	
0	None
1	Slightly wheezy
2	Moderately bad
3	Severe

Cough During Day	
0	None
1	Slight
2	Moderate
3	Severe

Breathless During Day	
0	Not breathless
1	More breathless than normal on vigorous exertion
2	Breathless on moderate exertion
3	Breathless on mild exertion

Sputum During Day	
0	None
1	Slight
2	Moderate
3	A lot

ASTHMA QUALITY OF LIFE QUESTIONNAIRE ©

Please complete **all** questions by circling the number that best describes how you have been during the **last 2 weeks as a result of your asthma.**

HOW **LIMITED** HAVE YOU BEEN **DURING THE LAST 2 WEEKS** IN THESE ACTIVITIES **AS A RESULT OF YOUR ASTHMA?**

	Totally Limited	Extremely Limited	Very Limited	Moderate Limitation	Some Limitation	A Little Limitation	Not at all Limited
1. STRENUOUS ACTIVITIES (such as hurrying, exercising, running up stairs, sports)	1	2	3	4	5	6	7
2. MODERATE ACTIVITIES (such as walking, housework, ardening, shopping, climbing stairs)	1	2	3	4	5	6	7
3. SOCIAL ACTIVITIES (such as talking, playing with pets/children, visiting friends/relatives)	1	2	3	4	5	6	7
4. WORK-RELATED ACTIVITIES (tasks you have to do at work*)	1	2	3	4	5	6	7

**If you are not employed or self-employed, these should be tasks you have to do most days.*

5. SLEEPING	1	2	3	4	5	6	7

HOW MUCH **DISCOMFORT OR DISTRESS** HAVE YOU FELT **DURING THE LAST 2 WEEKS?**

	A Very Great Deal	A Great Deal	A Good Deal	Moderate Amount	Some	Very Little	None
6. How much discomfort or distress have you felt over the last 2 weeks as a result of CHEST TIGHTNESS?	1	2	3	4	5	6	7

ASTHMA QUALITY OF LIFE QUESTIONNAIRE ©

IN GENERAL, **HOW MUCH OF THE TIME DURING THE LAST 2 WEEKS** DID YOU:

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	Hardly Any of the Time	None of the Time
7. Feel CONCERNED ABOUT HAVING ASTHMA?	1	2	3	4	5	6	7
8. Feel SHORT OF BREATH as a result of your asthma?	1	2	3	4	5	6	7
9. Experience asthma symptoms as a RESULT OF BEING EXPOSED TO CIGARETTE SMOKE	1	2	3	4	5	6	7
10. Experience a WHEEZE in your chest?	1	2	3	4	5	6	7
11. Feel you had to AVOID A SITUATION OR ENVIRONMENT BECAUSE OF CIGARETTE SMOKE?	1	2	3	4	5	6	7

HOW MUCH **DISCOMFORT OR DISTRESS** HAVE YOU FELT **DURING THE LAST 2 WEEKS?**

	A Very Great Deal	A Great Deal	A Good Deal	Moderate Amount	Some	Very Little	None
12. How much discomfort or distress have you felt over the last 2 weeks as a result of COUGHING?	1	2	3	4	5	6	7

IN GENERAL, **HOW MUCH OF THE TIME DURING THE LAST 2 WEEKS** DID YOU:

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	Hardly Any of the Time	None of the Time
13. Feel FRUSTRATED as a result of your asthma?	1	2	3	4	5	6	7
14. Experience a feeling of CHEST HEAVINESS?	1	2	3	4	5	6	7

ASTHMA QUALITY OF LIFE QUESTIONNAIRE ©

IN GENERAL, **HOW MUCH OF THE TIME DURING THE LAST 2 WEEKS** DID YOU:

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	Hardly Any of the Time	None of the Time
15. Feel CONCERNED ABOUT THE NEED TO USE MEDICATION for your asthma?	1	2	3	4	5	6	7
16. Feel the need to CLEAR YOUR THROAT?	1	2	3	4	5	6	7
17. Experience asthma symptoms as a RESULT OF BEING EXPOSED TO DUST?	1	2	3	4	5	6	7
18. Experience DIFFICULTY BREATHING OUT as a result of your asthma?	1	2	3	4	5	6	7
19. Feel you had to AVOID A SITUATION OR ENVIRONMENT BECAUSE OF DUST?	1	2	3	4	5	6	7
20. WAKE UP IN THE MORNING WITH ASTHMA SYMPTOMS?	1	2	3	4	5	6	7
21. Feel AFRAID OF NOT HAVING YOUR ASTHMA MEDICATION AVAILABLE?	1	2	3	4	5	6	7
22. Feel bothered by HEAVY BREATHING?	1	2	3	4	5	6	7
23. Experience asthma symptoms as a RESULT OF THE WEATHER OR AIR POLLUTION OUTSIDE?	1	2	3	4	5	6	7
24. Were you WOKEN AT NIGHT by your asthma?	1	2	3	4	5	6	7
25. AVOID OR LIMIT GOING OUTSIDE BECAUSE OF THE WEATHER OR AIR POLLUTION?	1	2	3	4	5	6	7

ASTHMA QUALITY OF LIFE QUESTIONNAIRE ©

IN GENERAL, **HOW MUCH OF THE TIME DURING THE LAST 2 WEEKS** DID YOU:

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	Hardly Any of the Time	None of the Time
26. Experience asthma symptoms as a RESULT OF BEING EXPOSED TO STRONG SMELLS OR PERFUME?	1	2	3	4	5	6	7
27. Feel AFRAID OF GETTING OUT OF BREATH?	1	2	3	4	5	6	7
28. Feel you had to AVOID A SITUATION OR ENVIRONMENT BECAUSE OF STRONG SMELLS OR PERFUME?	1	2	3	4	5	6	7
29. Has your asthma INTERFERED WITH GETTING A GOOD NIGHT'S SLEEP?	1	2	3	4	5	6	7
30. Have a feeling of FIGHTING FOR AIR?	1	2	3	4	5	6	7

HOW LIMITED HAVE YOU BEEN DURING THE LAST 2 WEEKS?

	Most Not Done		Several Not Done		Very Few Done		No Limitation
31. Think of the OVERALL RANGE OF ACTIVITIES that you would have liked to have done during the last 2 weeks? How much has your range of activities been limited by your asthma?	1	2	3	4	5	6	7

ASTHMA QUALITY OF LIFE QUESTIONNAIRE ©

HOW LIMITED HAVE YOU BEEN **DURING THE LAST 2 WEEKS?**

	Totally Limited	Extremely Limited	Very Limited	Moderate Limitation	Some Limitation	A Little Limitation	Not at all Limited
32. Overall, among ALL THE ACTIVITIES that you have done during the last 2 weeks, how limited have you been by your asthma?	**1**	**2**	**3**	**4**	**5**	**6**	**7**

DOMAIN CODE:

Symptoms: 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, 29, 30

Activity Limitations: 1, 2, 3, 4, 5, 11, 19, 25, 28, 31, 32

Emotional Function: 7, 13, 15, 21, 27

Environmental Stimuli: 9, 17, 23, 26

ASTHMA CONTROL QUESTIONNAIRE ©

Please answer questions 1 - 6.

Circle the number of the response that best describes how you have been during the past week.

1. On average, during the past week, how often were you **woken by your asthma** during the night?	0	Never
	1	Hardly ever
	2	A few times
	3	Several times
	4	Many times
	5	A great many times
	6	Unable to sleep because of asthma
2. On average, during the past week, how **bad were your asthma symptoms when you woke up** in the morning?	0	No symptoms
	1	Very mild symptoms
	2	Mild symptoms
	3	Moderate symptoms
	4	Quite severe symptoms
	5	Severe symptoms
	6	Very severe symptoms
3. In general, during the past week, how **limited were you in our activities** because of your asthma?	0	Not limited at all
	1	Very slightly limited
	2	Slightly limited
	3	Moderately limited
	4	Very limited
	5	Extremely limited
	6	Totally limited
4. In general, during the past week, how much **shortness of breath** did you experience because of your asthma?	0	None
	1	A very little
	2	A little
	3	A moderate amount
	4	Quite a lot
	5	A great deal
	6	A very great deal

ASTHMA CONTROL QUESTIONNAIRE ©

5. In general, during the past week, how much of the time did you **wheeze?**	0	Not at all
	1	Hardly any of the time
	2	A little of the time
	3	A moderate amount of the time
	4	A lot of the time
	5	Most of the time
	6	All the time
6. On average, during the past week, how many **puffs/inhalations of short-acting bronchodilator** (e.g. Ventolin/Bricanyl) have you used each day? *(If you are not sure how to answer this question, please ask for help)*	0	None
	1	1 - 2 puffs/inhalations most days
	2	3 - 4 puffs/inhalations most days
	3	5 - 8 puffs/inhalations most days
	4	9 - 12 puffs/inhalations most days
	5	13 - 16 puffs/inhalations most days
	6	More than 16 puffs/inhalations most days

To be completed by a member of the clinic staff

7. FEV_1 pre-bronchodilator:	0	> 95% predicted
	1	95- 90%
FEV_1 predicted:	2	89- 80%
	3	79- 70%
	4	69- 60%
FEV_1 % predicted:	5	59- 50%
(Record actual values on the dotted lines and score the FEV_1 % predicted in the next column)	6	< 50% predicted

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Amended Statistical Analysis Plan for Bayesian Analysis

Scott M. Berry, PhD., Donald A. Berry, PhD., and Ashish Sanil, PhD.
Berry Consultants, LLC
Version 3, August 20, 2008

This document describes the statistical details for the randomized clinical trial comparing the Alair® System to a sham control procedure.

Approved by:	**Signature**	**Date**
Scott M. Berry, PhD Berry Consultants, LLC.	SMB	8-20-2008
Donald A. Berry, PhD Berry Consultants, LLC.		
Ashish Sanil, PhD Berry Consultants, LLC.		

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Ashish Sanil, PhD Berry Consultants, LLC.	_____	AUGUST 20, 2008

Primary Outcome

The primary efficacy outcome will be the difference between study groups in the change between baseline and the average of the 6-, 9-, and 12-month follow-up Asthma Quality of Life Questionnaire (AQLQ) scores. The AQLQ score is a numeric score on a 7-point scale with higher scores representing better quality of life. The average of the 6-, 9-, and 12-month differences in the AQLQ score from baseline are referred to as the integrated AQLQ score.

Randomization

The randomization is 2:1 for Alair® System (treatment) to control.

Sample Size

The goal is for a sample size of 225 after subjects who withdraw or are lost-to-follow-up. This sample size results in an expectation of 150 treatment subjects and 75 control subjects. The study will accrue 250 subjects to make sure, after lost-to-follow-up, that there is a minimum of 225 evaluable subjects. During accrual, if the blinded results show an attrition rate larger than the expected 10% the study may continue enrolling up to a maximum of 300 subjects.

Final 12-month Analysis

Primary Statistical Analysis

Each subject will have a baseline, 3-month, 6-month, 9-month, and 12-month measurement of AQLQ. For subject i label these AQLQ results as X_{0i}, X_{3i}, X_{6i}, X_{9i}, and X_{12i}. Let Y_{3i}, Y_{6i}, Y_{9i}, and Y_{12i} be the change in AQLQ from baseline: $Y_{3i} = (X_{3i} - X_{0i})$, $Y_{6i} = (X_{6i} - X_{0i})$, $Y_{9i} = (X_{9i} - X_{0i})$, and $Y_{12i} = (X_{12i} - X_{0i})$. A positive change indicates an increase in the AQLQ, which is a positive outcome for a subject. The integrated AQLQ score for subject i is $Y_i = (Y_{6i} + Y_{9i} + Y_{12i})/3$.

We use the following Analysis of Covariance (ANCOVA) model that incorporates the baseline AQLQ score as a covariate

$$Y_i = \mu + \delta_{t(i)} + \beta\left(X_{0i} - \overline{X}_0\right) + \varepsilon_i \quad \text{with } t(i) \in \{T,C\}, \delta_c \equiv 0, \text{ and } \varepsilon \sim N(0,\sigma^2)$$

We calculate the posterior probability of superiority, π

$$\pi = \Pr[\delta_T > \tilde{0} \mid \text{Trial Results}].$$

We use independent $N(0,100^2)$ priors for the μ, δ_T and β, and $\sigma^2 \sim IG(0.01,100)$.

If the posterior probability, π, is larger than 0.964* then Alair will be considered superior to control.

* *This value must be selected to control the type I error of this design at no greater than 0.05.*

Lost to Follow-Up and Missing Data

In keeping with an intent-to-treat philosophy, the primary analysis is done on all randomized subjects administered at least one bronchoscopy. Those subjects who withdraw, are lost to follow-up (LTFU), or have missing data will be included in the analysis. If 2 or more of the 3 time periods for the integrated AQLQ are received the average of these two will be treated as the final integrated AQLQ value. Those subjects missing more than one of the 6-, 9-, and 12-month AQLQ values will be included using Bayesian multiple imputation. The same technique which is used for the predictive distribution in the interim analysis will be used to model these subjects.

Under this model, if one or more of the Y's are missing, then their distribution conditional on the observed Y's is also a normal distribution that is readily computed. This is the key idea behind the multiple imputation strategy employed here. If some of the Y-values are missing, we simulate values from their complete conditional distribution given other Y-values. For each of the Y-values (observed and possibly simulated), an analysis is conducted. The reported estimates are averaged over all these simulations, and thereby incorporate the error due to "missingness" in a natural and principled manner.

This is the primary analysis, but for sensitivity purposes, we conduct the following additional analyses.

1) We analyze only subjects with complete baseline, 6-, 9-, and 12-month AQLQ data.
2) For subjects with at least 1 of the 3 time periods that make up the integrated AQLQ (6-, 9-, and 12-months) we assume their partial integrated AQLQ score is in fact the true integrated AQLQ—a form of last-observation-carried-forward (LOCF).
3) Extreme cases will be presented to capture the range of possibilities. Specifically, we consider four cases:
   a) Assume all missing observations under treatment and control had the best possible prognosis
   b) Assume all missing observations under treatment and control had the worst possible prognosis
   c) Assume all missing observations under treatment had the best possible prognosis and all missing observations under the control had the worst possible prognosis
   d) Assume all missing observations under treatment had the worst possible prognosis and all missing observations under the control had the best possible prognosis.

We recognize that this is a very difficult problem—there is no right answer. We believe the Bayesian imputation approach is better than alternatives because it recognizes the variability present. LOCF or plugging in extreme values underestimates the variability present. We also recognize the Bayesian imputation can suffer from bias, as any technique can. For this reason we also present the sensitivity analysis described above.

Secondary Efficacy Endpoint Analyses

Secondary endpoint analyses will be conducted for the following efficacy endpoints:

1. Percentage of Symptom Free Days.
2. Symptom scores.
3. Morning Peak Expiratory Flow (amPEF).
4. Asthma Quality of Life Questionnaire (AQLQ) score (evaluations at 6-, 9- and 12-months), controlled for baseline AQLQ score.
5. Asthma Control Questionnaire (ACQ) Score.
6. Number of puffs of rescue medication used.
7. Percent Days rescue medication was used.
8. Forced Expiratory Volume in one second (FEV1).

With the exception of item (4) above, each analysis will assess the difference between Study groups in "Change from Baseline" for the particular endpoint at 6 and 12 months (6- and 12-months after last bronchoscopy session). Simple "difference of group-means" analyses will be conducted for each for each endpoint using non-informative Normal priors ($N(0,100^2)$) for the means, and non-informative inverse-gamma priors (IG(0.01,100)) for the variances.

The analyses for number 4 are done using the ANCOVA model defined in the primary analysis section. All analyses for secondary endpoints will be done on an LOCF basis.

Assessment of Site Heterogeneity

The testing of site heterogeneity will not be done from a Bayesian perspective. There are straightforward standard tests from a frequenist perspective. The ANOVA model (as described in the SAP) is utilized to aid in the acceptability, calculation, and ease of analysis. Should site heterogeneity become an issue (significant heterogeneity exits) then a Bayesian ANCOVA hierarchical models would be utilized to model the site heterogeneity.

Adverse Events

Inferential analyses of adverse events (AEs) are not done as the default. If inferential analyses of AEs become warranted (see SAP) then the following Bayesian model is used. Let the probability of an AE for a subject, for each treatment group be π_T and π_C, respectively. The following independent prior distributions are assumed

$$\pi_T, \pi_C \sim \text{Beta}(0.1, 0.1).$$

The posterior probability of an elevated rate of AEs in the treatment (control) group are presented.

$$\Pr(\pi_T > \pi_C \mid \text{data}).$$

Early Analysis Looks (Prior to the Final Bayesian Analysis which Incorporates Baseline AQLQ as a Covariate)

Two interim analyses take place after all subjects have been accrued. These interim analyses are for the purpose of an early claim of success. These analyses take place when 225

subjects reach their 6-month follow-up visit and when 225 subjects reach their 9-month visit. All data available at the time of these analyses will be included in the interim look. If the predictive probability of trial success exceeds 0.99 then an immediate claim of success will be made.

At the time of the interim analysis there will be subjects who have completed 12 months, 9 months, 6 months, as well as subjects who have 3-month results and subjects who have enrolled but don't have any interim results. We employ Bayesian predictive distributions for the imputation of incomplete results. We use a regression model with a normal distribution for the prediction of the next follow up time, based on the most recent observation. Details of the statistical modeling for the predictive distributions are presented in the next section.

If the result of the study is immediate success a PMA will be filed, but all subjects will be followed to the end of the study.

Two safety interim analyses are planned, the first of which will occur after 225 subjects have reached their 6-week follow-up, and the second of which will occur after 225 subjects have reached their 3 month follow-up. These analyses will be done to get the logistics of an interim analysis straight, refine the timing, and for monitoring the safety of the Alair device. There are no decisions planned at these times and thus these provide logistical practice and the ability to address safety concerns. These looks safety/practice interim analyses have no effect on the operating characteristics of the design.

Predictive Probabilities

This section describes the calculation of the predictive probabilities. At each interim analysis there are subjects with differential information. There are 5 types of subjects: those who have completed 12 months, those who have 9-month interim results, 6-month interim results, 3-month interim results, and those with no interim results. For subjects with incomplete information we find the predictive distribution for these missing results. This modeling enables a predictive distribution for the integrated result of each subject. We use the same model within each treatment group, but treat them separately by assuming different parameters for each treatment.

The following statistical modeling is used to predict missing values. The subject indicator i is suppressed and t denotes the treatment: $t = 1$ (Control), 2 (Alair treatment).

$$[\, Y_3 \,] \sim N(\alpha_{t1}, \tau_1^2)$$
$$[\, Y_6 \mid Y_3 \,] \sim N(\alpha_{t2} + \beta_{t2} Y_3, \tau_2^2)$$
$$[Y_9 \mid Y_6 \,] \sim N(\alpha_{t3} + \beta_{t3} Y_6, \tau_3^2)$$
$$[Y_{12} \mid Y_9 \,] \sim N(\alpha_{t4} + \beta_{t4} Y_9, \tau_4^2)$$

The intercepts α_{t1}, α_{t2} , α_{t3} and α_{t4} have the following prior distributions:

$$\alpha_{tj} \sim N(0, \tau_j^2) \text{ for } t = 1,2 \text{ and } j = 1,\ldots,4.$$

The slopes β_{t2} , β_{t3} and β_{t4} have the following prior distributions:

$$\beta_{tj} \sim N(0.75, \tau_j^2) \text{ for } t = 1,2 \text{ and } j = 2,3,4.$$

These prior distributions are constructed to have one observation worth of weight in the posterior distribution (variance τ^2). These distributions carry information that neighboring observations are likely to be correlated, but the prior is overwhelmed by the data.

The variance components, τ_1^2, τ_2^2, τ_3^2, and τ_4^2 have inverse-gamma priors with parameters 3 and 0.5 (the mean of each τ^2 is 1). This distribution likewise is weak, but carries slight information that the τ^2 are likely to be near 1.

Subjects who have observations at a follow-up time are used to update the posterior distribution for the parameters for the respective transitional model.

A sampling approach is used to calculate predictive distributions. A Markov Chain approach is used to simulate an observation from the joint distribution of α_{11}, α_{12}, α_{13}, α_{14}, α_{21}, α_{22}, α_{23}, α_{24}, β_{12}, β_{13}, β_{14}, β_{22}, β_{23}, β_{24}, τ_1^2, τ_2^2, τ_3^2, and τ_4^2. Each missing Y_j is simulated, serially, conditioning on the simulated values from the posterior. These simulated Y_j values create a simulated integrated AQLQ, Y. The collection of Y's from each subject (or possible subject) is then an observation from the predictive distribution of the end of trial results. By repeating this process and simulating many trials (1,000 to 10,000) the predictive probability of trial success is the proportion of simulated trials in which a successful claim of superiority is made.

Operating Characteristics

This section presents the operating characteristics of the design. The details for simulating subjects for the trial are presented in the next section. Table 1 presents the operating characteristics for the design. The ε column presents the difference between the treatment and control. This treatment effect is assumed to be present at each of the interim follow-up visits, and thus is also the treatment difference for the integrated AQLQ. The $\varepsilon = 0$ row corresponds to the null hypothesis. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of an early claim of success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.0394	0.014	0.0055	0.0084
0.10	0.176	0.077	0.026	0.050
0.20	0.456	0.269	0.115	0.154
0.30	0.748	0.555	0.295	0.260
0.40	0.937	0.827	0.559	0.268
0.50	0.989	0.955	0.787	0.168
0.60	0.999	0.994	0.934	0.060

Table 1: The ε column presents the difference mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of

success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 10000 simulations were done.

For example, in the null hypothesis case there is a 0.039 probability of the trial resulting in a claim of superiority. In the case of $\varepsilon = 0.50$ there is a 0.989 probability of a successful claim of superiority (power), with a 0.955 probability of an early claim of success.

Simulation Details

In this section the details of the simulation process are presented. We make a series of different assumptions in simulating the subjects. Many of these assumptions affect the simulations but have no impact on how the results of the trial will be analyzed. This includes the assumptions made to simulate subjects.

We simulate 225 subjects. The expectation is 250 subjects will be accrued with at least 225 available with complete follow-up information. The accrual rate is assumed to be (per month): 5, 4, 9, 7, 9, 18, 20, 20, 20, 30, 30, 30, 30, 30, 38, and then 25 every month until the study is completed.

Subjects are simulated as follows:

$$[Y_3] \sim N(\mu_{t1}, \sigma_1^2)$$

$$[Y_6|Y_3] \sim N(\mu_{t2} + (Y_3-\mu_{t1})(\rho_2\sigma_2^2/\sigma_1^2), \sigma_2^2(1-\rho_2^2))$$

$$[Y_9|Y_6] \sim N(\mu_{t3} + (Y_6-\mu_{t2})(\rho_3\sigma_3^2/\sigma_2^2), \sigma_3^2(1-\rho_3^2))$$

$$[Y_{12}|Y_9] \sim N(\mu_{t4} + (Y_9-\mu_{t3})(\rho_4\sigma_4^2/\sigma_3^2), \sigma_4^2(1-\rho_4^2))$$

For the default simulation presented in the previous section the unconditional means of each Y_j are $\mu_{11}=\mu_{12}=\mu_{13}=\mu_{14}= 0$ (for the controls) and $\mu_{11}=\mu_{12}=\mu_{13}=\mu_{14}= \varepsilon$ (for the treatments). The unconditional variances are $\sigma_1^2 =0.90^2$, $\sigma_2^2=1$, $\sigma_3^2=1$, $\sigma_4^2=1.05^2$. The correlation coefficients are assumed to be $\rho_2=0.50$, $\rho_3=0.60$, $\rho_4=0.70$.

The parameters for each treatment group were selected from the AIR Trial (protocol: 0602-20) data set. Exact values from the data set were not selected because of the small sample sizes, but the general structure is maintained. The parameter ε is the mean difference between the treatment and control groups. We perform a sensitivity analysis to some of these assumptions to make sure the behavior of the trial is robust to these assumptions. The results are in the Sensitivity section.

We run 10000 simulations for the null case. The cut-off for superiority in the primary analysis, 0.964, is selected by trial-and-error in order to maintain a 0.05 type I error.

Sensitivity

In this section we present 9 scenarios in which the assumptions for simulating the data are altered. In each of these cases we present operating characteristics to compare to the primary operating characteristics.

Scenario A: In this scenario we assume that the effect of the treatment (ε) is not constant throughout the 6 months of integration. The treatment effect is 0.5ε, ε, and 1.5ε respectively at the 6-month, 9-month, and 12-month follow-up visits. The average of these creates a mean for the integrated AQLQ which is ε. The mean for the 3-month value is assumed to be $0.5\ \varepsilon$.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.0394	0.014	0.0055	0.0084
0.10	0.165	0.060	0.021	0.039
0.20	0.464	0.263	0.127	0.136
0.30	0.742	0.519	0.290	0.229
0.40	0.923	0.828	0.554	0.274
0.50	0.995	0.967	0.813	0.154
0.60	0.998	0.991	0.926	0.065

Table 2A: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario B: In this scenario the effect of the treatment (ε) is not constant throughout the 6 months of integration. The treatment effect is 1.5ε, ε, and 0.5ε, respectively, at the 6-month, 9-month, and 12-month follow-up visits. The average of these creates a mean for the integrated AQLQ which is ε. The mean for the 3-month value is assumed to be 1.5ε.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.0394	0.014	0.0055	0.0084
0.10	0.165	0.061	0.021	0.040
0.20	0.456	0.272	0.123	0.149
0.30	0.764	0.573	0.297	0.276
0.40	0.930	0.814	0.561	0.253
0.50	0.987	0.960	0.810	0.150
0.60	1	0.996	0.939	0.057

Table 2B: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario C: In this scenario a faster accrual rate is used. The assumed accrual rate is increased 50% for this scenario.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.043	0.007	0.003	0.004
0.10	0.165	0.066	0.023	0.043
0.20	0.465	0.246	0.082	0.164
0.30	0.733	0.522	0.237	0.285
0.40	0.936	0.792	0.428	0.364
0.50	0.993	0.959	0.703	0.256
0.60	1	0.989	0.865	0.124

Table 2C: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario D: In this example a slower accrual rate is assumed. The accrual numbers are multiplied by 0.5 to create a much slower accrual rate.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.033	0.020	0.010	0.010
0.10	0.155	0.093	0.053	0.040
0.20	0.459	0.316	0.185	0.131
0.30	0.764	0.647	0.473	0.174
0.40	0.934	0.890	0.763	0.127
0.50	0.993	0.979	0.940	0.039
0.60	0.999	0.998	0.984	0.014

Table 2D: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario E: In this scenario the follow-up at 3-months is assumed to be independent from the 6-month AQLQ score. Therefore, $\rho_2 = 0$ is assumed.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.047	0.014	0.006	0.008
0.10	0.168	0.060	0.023	0.037
0.20	0.467	0.204	0.078	0.126
0.30	0.750	0.512	0.255	0.257
0.40	0.928	0.785	0.508	0.277
0.50	0.993	0.952	0.781	0.171
0.60	0.999	0.992	0.932	0.060

Table 2E: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario F: In this scenario the correlations between time points are increased. The following values are assumed: $\rho_2 = 0.70$, $\rho_3 = 0.80$, $\rho_4 = 0.90$.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.045	0.020	0.005	0.015
0.10	0.160	0.085	0.034	0.051
0.20	0.392	0.254	0.123	0.131
0.30	0.678	0.507	0.307	0.200
0.40	0.872	0.772	0.565	0.207
0.50	0.971	0.926	0.801	0.125
0.60	0.995	0.987	0.939	0.048

Table 2F: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario G: In the scenario the same mean structure is used, but the correlations between time points are all 0; $\rho_1 = 0$, $\rho_2 = 0$, $\rho_3 = 0$, $\rho_4 = 0$.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.046	0.015	0.006	0.009
0.10	0.302	0.110	0.047	0.063
0.20	0.728	0.444	0.203	0.241
0.30	0.954	0.778	0.466	0.312
0.40	1	0.967	0.772	0.195
0.50	1	0.998	0.944	0.054
0.60	1	1	0.987	0.013

Table 2G: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario H: In this scenario each of the variances are doubled; $\sigma_1^2 = 2(.90^2)$, $\sigma_2^2 = 2(1)$, $\sigma_3^2 = 2(1)$, $\sigma_4^2 = 2(1.05^2)$.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.046	0.014	0.005	0.009
0.10	0.110	0.110	0.021	0.029
0.20	0.289	0.144	0.054	0.090
0.30	0.515	0.288	0.126	0.162
0.40	0.686	0.495	0.247	0.248
0.50	0.871	0.712	0.426	0.286
0.60	0.962	0.862	0.612	0.250

Table 2H: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Scenario I: In this scenario each of the variances are halved; $\sigma_1^2 = 0.5(.90^2)$, $\sigma_2^2 = 0.5(1)$, $\sigma_3^2 = 0.5(1)$, $\sigma_4^2 = 0.5(1.05^2)$.

ε	P(S)	P(Early)	P(225@6)	P(225@9)
0	0.049	0.015	0.005	0.010
0.10	0.267	0.126	0.048	0.078
0.20	0.693	0.486	0.216	0.270
0.30	0.968	0.884	0.632	0.252
0.40	0.998	0.987	0.902	0.085
0.50	1	1	0.989	0.011
0.60	1	1	0.999	0.001

Table 2I: The ε column presents the difference in mean integrated AQLQ between the treatment and control. The "P(S)" column reports the probability that the trial results in a successful claim of superiority. The "P(Early)" column reports the probability of immediate success. The probability of success on each of the two interim analyses are reported in the "P(225@6)" and "P(225@9)" columns. For each scenario 1000 simulations were done.

Changes to the Original Bayesian Analysis Plan Version 2, Dated December 12, 2006

The original Bayesian Analysis Plan has been amended as follows:

1. The baseline AQLQ value was recognized to be an important covariate in the analysis of change in AQLQ. Thus, all Bayseian analyses of AQLQ will be adjusted for baseline AQLQ (see letter from FDA dated June 6, 2008 (G050082/S030) and letters to FDA dated July 1, 2008 (G050082/S032 and S033)).

2. Modifications have been made to the terminology used to describe the parameters of the primary analyses. Previously, they were expressed in terms of a difference between mean effects (μ_a-μ_b). In the updated Bayesian Analysis Plan they are parameterized by $\delta = (\mu_a-\mu_b)$. This mathematically equivalent formulation allows consistentency in presenting the results of the primary and repeated-measures analyses.

3. Interim (blinded) data indicate that the amount of missing data related to the primary efficacy endpoints appear to be almost negligible. The missingness rate is further diminished for analysis purposes if one takes the non-model-based imputation prescription in the Lost to Follow-Up And Missing Data section. In light of this, we forgo one of the items -- a regression analysis with covariates -- that was originally proposed as a part of the sensitivity analysis. (A regression with covariates was originally planned to check if it informed the imputation procedure in any significant way. This was a relatively delicate and complex procedure to uncover potential subtle biases in the imputation methods.) With the exception of this regression analysis, we intend, as in the original plan, to conduct a sensitivity assessment using the analyses listed in the **Lost to Follow-Up And Missing Data** section.

4. In the secondary endpoint analyses of AQLQ a subset evaluation with restricted Baseline AQLQ was proposed. Since all AQLQ analyses include baseline AQLQ as a covariate only descriptive statistics will be provided by treatment arm for the subsets AQLQ < 2, 2 ≤ AQLQ < 3, 3 ≤ AQLQ < 4, 4 ≤ AQLQ < 5, 5 ≤ AQLQ < 6, and 6 ≤ AQLQ.

5. The analysis of site-to-site variability will be performed using classical ANCOVA modeling for ease of computation and standardization of this testing (See section 6.1). Bayesian ANCOVA hierarchical modeling will be employed should site-to-site heterogeneity become an issue.

6. Predictive probabilities of a difference between treatment arms will be presented for adverse events whose incidence rates are 3% or higher in either treatment arm. In cases where inferential analyses of AEs are required, the posterior probability that the AE rate for the treatment group exceeds the AE rate of the control group will be reported (the posterior probability that the control AE rate exceeds the treatment AE rate is just 1 minus the reported posterior probability).